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Hydrolyzable tannins from hydroalcoholic extract from *Poincianella pluviosa* stem bark and its wound-healing properties: Phytochemical investigations and influence on in vitro cell physiology of human keratinocytes and dermal fibroblasts

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ABSTRACT

Extracts from *Poincianella pluviosa* stem bark are used in traditional medicine of South America for its wound healing properties. For validation of this traditional use and for rationalizing a potential pharmaceutical development towards standardized preparations bioassay-guided fractionation of EtOH–water (1:1 v/v) extract (crude extract, CE) of *P. pluviosa* bark was performed. HaCaT keratinocytes cell line and human primary dermal fibroblasts (pNHDF) were used as in vitro systems. Significant stimulation of mitochondrial activity was found for CE on both cell types, which caused a strong increase of cell proliferation of keratinocytes. Fractionation of CE over Sephadex LH20 revealed two inactive fractions (FA and FB) and an active fraction FC, which was further fractionated by MPLC into 4 subfractions. Subfraction FC1 increased mitochondrial activity and proliferation of keratinocytes and dermal fibroblasts in a dose dependent manner (10 to 100 μg/mL) and did not show necrotic cytotoxicity on keratinocytes (LDH release assay). FC1 was investigated by ESI–MS/MS and solid-state ^{13}C NMR which confirmed the presence of various polyphenols and hydrolyzable tannins. MS studies suggest the presence of pyrogallol (**1**), gallic acid (**2**), gallic acid methyl ester (**3**), ellagic acid (**4**), corilagin (**5**), 1,4,6-tri-*O*-galloyl-glucose (**6**), tellimagrandin I (**7**), 1,2,3,6-tetra-*O*-galloyl-glucose (**8**), mallotinic acid (**9**), tellimagrandin II (**10**), 1,2,3,4,6-penta-*O*-galloyl-glucose (**11**), geraniin (**12**), and mallotusinic acid (**13**).

1. Introduction

Wounds have a tremendous impact on the healthcare economy and intensified research for improvement of wound-healing is still a major goal. While in Europe and U.S. acute wounding is mostly treated by systematic disinfection and surgical procedures, less developed countries rely on the topical application of wound-healing plant extracts or natural compounds [1]. On the other side chronic wounds still represent a major health burden and drain on healthcare resources in the developed and undeveloped countries [2]. In many cases this plant-based preparations are based on traditional local medicine. The treatment and healthcare by using plant extracts with wound healing properties is inexpensive and can be managed easily by basically trained health workers [1]. Most of these medicinal plants have been used for a long time and are assessed to be cheaper and safer than

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isolated or chemical synthesized active compounds. Therapeutic strategies for chronic wounds have been published and are implemented in clinical therapy in developed countries but in many cases the success of such therapies is still very limited and many patients are suffering from chronic wounds over long time periods. In U.S. nearly 6 million people are estimated to suffer from unhealed wounds [3], and the cost of this problem reaches about ten billion dollars annually [4]. The skin forms a barrier that protects the body from intentional or accidental damage. Skin wounds are essentially the disruption of functional continuity of cells and tissues of the skin at the site of injury, and can be caused by physical, chemical, microbiological or immunological insults, which can compromise its function [5]. The integrity of the tissue can be reestablished by the wound-healing process, which is complex and involves several biological events, including vascular and cellular changes, epithelial proliferation, collagen synthesis and deposition, fibroblast proliferation and migration, revascularization, and wound contraction. The normal process involves three stages: inflammation, proliferation, and tissue remodeling. However, this process may be impaired and the healing time extended by some factors like severity and type of wounds, microbial infection, conditions of health, medications, sex, age, stress situations, immunity status, obesity and environment around the site of the wounds.

The need to search for compounds that have evident ability to accelerate the healing process is obvious. Natural products can play an important role in this sense, acting in the stimulation of cellular proliferation and differentiation of keratinocytes [6,7] and dermal fibroblasts [8], on induction of collagen synthesis [7] and improvement of innate immune response in the wound area. Especially polyphenols and polysaccharides have been published to act specifically on the relevant skin cells [9–11]. The research for new products is important for discovering new lead compounds.

The genus *Caesalpinia* consists worldwide of about 500 species [12] and many medical uses of species from the genus are related to antimalarial [13], antiulcer [14], anti-inflammatory [15], or wound healing activity [16,17]. *Poincianella pluviosa* (DC.) L.P. Queiroz is native from Brazil, commonly known as “sibipiruna” or “false Brazil wood”, and is also reported under its synonyms *Caesalpinia peltophoroides* (Benth.), *Caesalpinia pluviosa* DC., and *C. pluviosa* var. *peltophoroides* (Benth.) GP Lewis [18]. The species is found in Brazil primarily in the Atlantic Forest and Pantanal region [19] and is widely used in afforestation of Brazilian cities. Beside the use of this plant in Brazil for wound healing the bark of *P. pluviosa* has been described also in Bolivia for treatment of dysentery [13]. Recently, it has been shown that an extract of *P. pluviosa* showed an antimalarial activity [20]. Concerning the phytochemical composition of the species presence of sterols, tannins, flavonoids and saponins as determined by colorimetric group determinations has been confirmed, while alkaloids were found to be absent [21]. The essential oil of the flowers was shown to be dominated by long chain alkanes [22]. The flowers are described to contain brevifoincarboxylate, 5-hydroxymethylfurfural, luteolin, gallic acid, gallic acid ethyl ester, sitosterol, glucosylated sitosterol and squalene [23]. From the stem bark of *C. pluviosa* gallic acid, gallic acid ethyl ester, rhuschalcone VI, lupeol, betulinic acid and stigmasterol have been isolated [24].

The aim of the following study was the evaluation of a hydroalcoholic extract (CE) from the stem bark of *P. pluviosa* on potential wound healing activity by determination of its influence on cell in vitro physiology of human skin cells. Especially viability and proliferation of human keratinocytes as key players of human epidermis were to be investigated besides the activity of the extract and its fractions against dermal fibroblasts, representing the major cells from the skin dermis. Additionally, fractions obtained were to be investigated phytochemically by ESI–MS/MS and solid-state ^{13}C NMR analysis.

2. Materials and methods

2.1. General experimentation procedures

If not stated otherwise all chemicals were purchased from Sigma (Deisenhofen, Germany) or VWR (Darmstadt, Germany). Bark of *P. pluviosa* (DC.) L.P. Queiroz was collected at the Universidade Estadual de Maringá Campus, Maringá, Brazil and identified by Prof. Dr. Cássia Mônica Sakuragui. Voucher species are deposited in the herbarium of the Universidade Estadual de Maringá and Universidade Federal do Rio de Janeiro under numbers 12492 HUEM and RFA 39925, respectively.

2.2. Extraction and fractionation (Fig. 1)

Dried and milled bark material (2 kg) was extracted with 20 L of EtOH–water (1:1 v/v) by Ultra-Turrax® UTC115KT (Ika® Works, Wilmington, NC) for 3 × 5 min at 40 °C. The resulting extract was filtered (Mesh 0.130), concentrated under reduced pressure and lyophilized to yield 226 g (11.3%, related to the starting material) of the crude extract, named as CE. CE (3 × 50 g) was partitioned ten times between ethyl acetate (EtOAc):water (1:1 v/v; 3 × 1 L). After separation of the two phases the respective solvents were removed under reduced pressure and the extracts were lyophilized to yield the EtOAc-fraction (EAF, 23.70 g) and H_2O-fraction (WF, 120.60 g). 10 g of EAF was dissolved in ethanol 50% (v/v) and fractionated on Sephadex® LH-20 (Pharmacia Biotech AB, Uppsala, Sweden, 650 × 50 mm) using a step gradient of 1500 mL ethanol 50% (FA, 2.95 g) and 2200 mL ethanol 100% (FB, 1.04 g), and FC (5.43 g) was obtained by adding 2000 mL methanol 50% (0.3 g), 2000 mL methanol 100% (0.5 g), and 3000 mL acetone 70% (4.63 g). 200 mg of fraction FC was fractionated on MCI-Gel® CHP-20P stationary phase (Mitsubishi Kasei Corporation, Tokyo, Japan, 75–150 μm, 450 Å, 500 × 25 mm) by using a linear gradient of methanol and water as mobile phase (20% MeOH ⇒ 100% over 120 min at a flow rate of 5 mL/min) with a Waters 510 HPLC pump. This chromatographic separation yielded 4 subfractions FC1 (225 mL, 90.12 mg), FC2 (30 mL, 42.33 mg), FC3 (55 mL, 28.50 mg), and FC4 (215 mL, 21.71 mg).

All subfractions were monitored by analytical TLC on silica gel 60 F_{254} aluminium sheets (10 × 20 cm) using ethylacetate: water:formic acid (90:5:5; v/v/v) as mobile phase; detection was performed at daylight, λ = 366 and 254 nm, and after spraying with $FeCl_3$ (2% in ethanol). The subfractions were

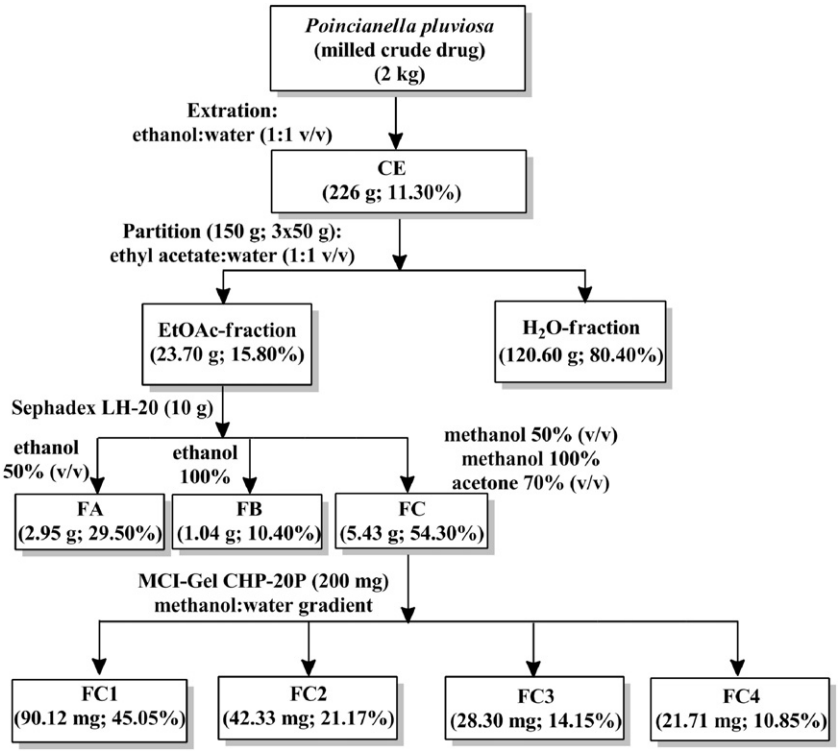

Fig. 1. Bioassay-guided fractionation of EtOH–water extract from *Poincianella pluviosa*.

combined based on the composition and pattern on the TLC. The respective solvents were removed under reduced pressure and the fractions were lyophilized. The total polyphenol and total tannin contents were determined using a modified Folin–Ciocalteu method [25,26].

2.3. Solid-state ^{13}C NMR and ESI–MS/MS

NMR spectra were obtained on a NMR Varian Mercury Plus 300BB spectrometer (Varian, USA) probe CPMAS 7 mm, a frequency of 75.45 MHz for ^{13}C, using FC1 in solid state. Each proton preparation pulse was 50 μs at 90°, with spun of 100 Hz and followed by 1000 μs of data acquisition and 1 s of recovery delay. Transient sequence was 7168. The spectra of the compounds were analyzed and compared to literature data.

Micromass Quattro *micro*™ API benchtop triple quadrupole mass spectrometer equipped with a Z-spray electrospray ionization (ESI) source (Waters, Milford, MA, USA) operating in negative mode was used. MassLynx™ software (version 4.0, Waters, Milford, MA, USA) was used for data acquisition and processing. Sample containing 500 ng/mL of FC1 was directly injected into MS source. Capillary voltage was set to 3.0 kV, cone to 35 V, and extraction cone to 2 V. The source temperature was 100 °C, desolvation temperature was 250 °C, cone gas flow was 50 L/h, and desolvation gas flow was 500 L/h. The scan time was 0.3 s with an interscan time of 0.02 s. Argon was used as a collision gas (collision energy from 15 to 30 eV). Preliminary analysis of MS was carried out using full scan, and the compounds were identified by MS/MS fragments.

2.4. HaCaT keratinocytes and fibroblasts in vitro cell culture

Ethical Committee of University of Münster, Germany (acceptance #2006-117-f-S) approved the studies. Primary natural dermal fibroblasts (pNHDF) were obtained from surgical resectates of Caucasian subjects by the University Hospital of Münster (Department of Dermatology and Paediatrics, Münster, Germany). The isolation and propagation of dermal fibroblasts from the dermis were performed according to [9]. HaCaT keratinocytes were provided by Prof. Dr. Norbert E. Fusenig (DKFZ, Heidelberg, Germany) and cultivated in a DMEM high glucose medium, supplemented with FCS (10%), penicillin/streptomycin solution (1%), glutamine (1%) and non-essential amino acids (1%) (PAA, Pasching, Austria). pNHDF and HaCaT keratinocytes were cultivated at 37 °C, 5% CO_2 and 37 °C, 8% CO_2, respectively.

Functional testing was performed with cells in a FCS-free medium (HaCaT keratinocytes: MCDB 153 complete, Biochrom, Berlin, Germany; pNHDF: MEM high glucose, SerEx® (10%) and L-glutamine (1%), PAA Pasching, Austria). In vitro tests were performed concerning mitochondrial activity by the MTT test [27], mitogenic cell proliferation rate by the BrdU incorporation assay [28], and release of lactate dehydrogenase (LDH) [8] against untreated control and 5% FCS treated cells as positive control [29].

2.5. Statistical analysis

Software Statistica® 8.0 (StatSoft Copyright, Inc. 1984–2007) was used for statistical analysis. Significant differences

were determined by the analysis of variance unilateral (one-way ANOVA-Tukey) test for multiple comparisons, and considered $p < 0.05$ (*) as significant.

3. Results and discussion

For in vitro investigation of a potential activity of *P. pluviosa* bark extract on human skin cells HaCaT keratinocytes and primary natural human dermal fibroblasts (pNHDF) were used to investigate the influence of an EtOH–water extract (1:1) (CE) on cell physiology. While cellular viability was evaluated by MTT assay [27], cellular proliferation was quantified by BrdU incorporation ELISA [28]. CE stimulated cell viability and mitochondrial activity of keratinocytes as well as of pNHDF in the concentration range of 10 to 50 μg/mL (Table 1).

It was interesting that CE also stimulated the cellular proliferation of keratinocytes, while in contrast the proliferation of dermal fibroblasts was dramatically decreased by CE in a concentration-dependent manner (Table 1). This contradictory result for the two different cell types can only be correlated with a potential triggering effect of CE towards cellular differentiation of dermal fibroblasts, which could explain the stop of proliferation by increased metabolic activity, but decreased mitogenic proliferation.

Bioassay-guided fractionation was performed by chromatography of CE on Sephadex LH20, using different mobile phases. Total fractions obtained from the different solvents were named as FA, FB, and FC and investigated concerning their respective activity against the skin cells. While FA and FB were inactive or exerted slightly toxic effects against keratinocytes and dermal fibroblasts within MTT assay, fraction FC showed strong stimulation of viability for both cell types (Table 1).

FC was further fractionated by reversed phase chromatography on MCI gel using a MeOH–water gradient, which resulted in the isolation of four subfractions FC1 to FC4. In vitro cell testing indicated FC1 and FC2 to be strong stimulators of mitochondrial activity of keratinocytes (Fig. 2A) and dermal fibroblasts (Fig. 2B), while FC3 and FC4 were inactive or even toxic.

Investigation of FC1 and FC2 on the influence on cellular proliferation of HaCaT keratinocytes (Fig. 2C) indicated FC1 to induce mitogenic cell proliferation significantly at 10 to 25 μg/mL, while FC2 inhibited cell proliferation. Therefore FC1 is assessed as a strong stimulant of epidermal proliferation, while FC2 seems to trigger cells into cellular differentiation. Quantification of LDH release from keratinocytes by FC1 revealed no significant differences to the respective control groups, indicating the absence of necrotic cell activity in the range from 1 to 100 μg/mL (data not shown). From this bioactivity-guided fractionation it can be concluded that CE, FC, and FC1 are able to influence the cell physiology of skin cells significantly. It seems interesting that proliferation (but not the viability!) of fibroblasts was decreased by the extract CE, while the proliferation of keratinocytes was significantly increased. This phenomenon can be interpreted in a way that the extract stops the mitogenic cell cycle of NHDF but triggers the cells into a stage of higher metabolic activity by induction of cellular differentiation, e.g. for production and secretion of polymers for the formation of the extracellular matrix. Several recent publications indicate similar effects of natural products against the two different cell types of the skin [7,11] and showed that keratinocytes and fibroblasts probably due the existence of different cellular targets act completely different on exogenous stimulus.

To obtain a deeper insight into the chemical composition of the active fractions analytical studies were performed. The polyphenol content of the EtOH–water extract CE of *P. pluviosa* was determined to be 22.7% (w/w) by UV-spectrometry [26] while FC1 contained 54.1% ± 1.8 (mean ± SD) of total phenol content and 54.1% ± 1.8 of total tannin content.

FC1 was investigated by solid-state ^{13}C NMR spectroscopy (Fig. 3). The respective signals indicate the presence of aromatic *ortho*-orientated CH carbons at C_2 and C_3 at 111 ppm. At 143 ppm hydroxyl groups relative to C_3 and C_5, C_1, and C_4-OH at 121 and 135 ppm, respectively, were found [30]. A carboxyl group found at 168 ppm is characteristic for gallic acid [30], a precursor for some hydrolyzable and condensed tannins. Signals typical for lignin (153–151 ppm, relative to C-3/5

Table 1
Influence of CE and subfractions FA, FB and FC on cellular dehydrogenase activity (MTT assay) and mitogenic cell proliferation (BrdU assay) of HaCaT keratinocyte cell line and primary dermal fibroblasts (pNHDF) at 1, 10, 25, 50 and 100 μg/mL. The results are expressed as mean ± SD. Values are mean of 3 independent experiments with n = 8 replicates. NC: untreated negative control; PC: positive control (5% FCS); n.p.: not performed; *$p < 0.05$ compared to NC.

Cell viability (MTT assay)								
Fraction	Cell type	NC	PC	1	10	25 μg/mL	50	100
CE	HaCaT	100 ± 15	125 ± 05*	98 ± 12	110 ± 5*	138 ± 11*	124 ± 10*	105 ± 11
	pNHDF	100 ± 19	133 ± 10*	94 ± 10	110 ± 9*	134 ± 9*	120 ± 9*	97 ± 9
FA	HaCaT	100 ± 17	206 ± 14*	76 ± 6*	89 ± 12*	67 ± 6*	n.p.	n.p.
	pNHDF	100 ± 13	135 ± 16*	n.p.	71 ± 11*	86 ± 16*	89 ± 18*	75 ± 20*
FB	HaCaT	100 ± 08	206 ± 14*	100 ± 10	103 ± 08	74 ± 10	n.p.	n.p.
	pNHDF	100 ± 09	163 ± 6*	n.p.	89 ± 8*	83 ± 9*	80 ± 10*	77 ± 04*
FC	HaCaT	100 ± 12	189 ± 16*	61 ± 13*	95 ± 10	120 ± 13*	158 ± 09*	189 ± 10*
	pNHDF	100 ± 16	163 ± 6*	n.p.	118 ± 05*	162 ± 6*	158 ± 8*	139 ± 9*
Cell proliferation (BrdU assay)								
Fraction	Cell type	NC	PC	1	10	25 μg/mL	50	100
CE	HaCaT	100 ± 10	110 ± 3*	85 ± 3*	99 ± 7	126 ± 8*	147 ± 7*	119 ± 10*
	pNHDF	100 ± 13	259 ± 9*	71 ± 13*	58 ± 3*	19 ± 13*	14 ± 6*	12 ± 8*

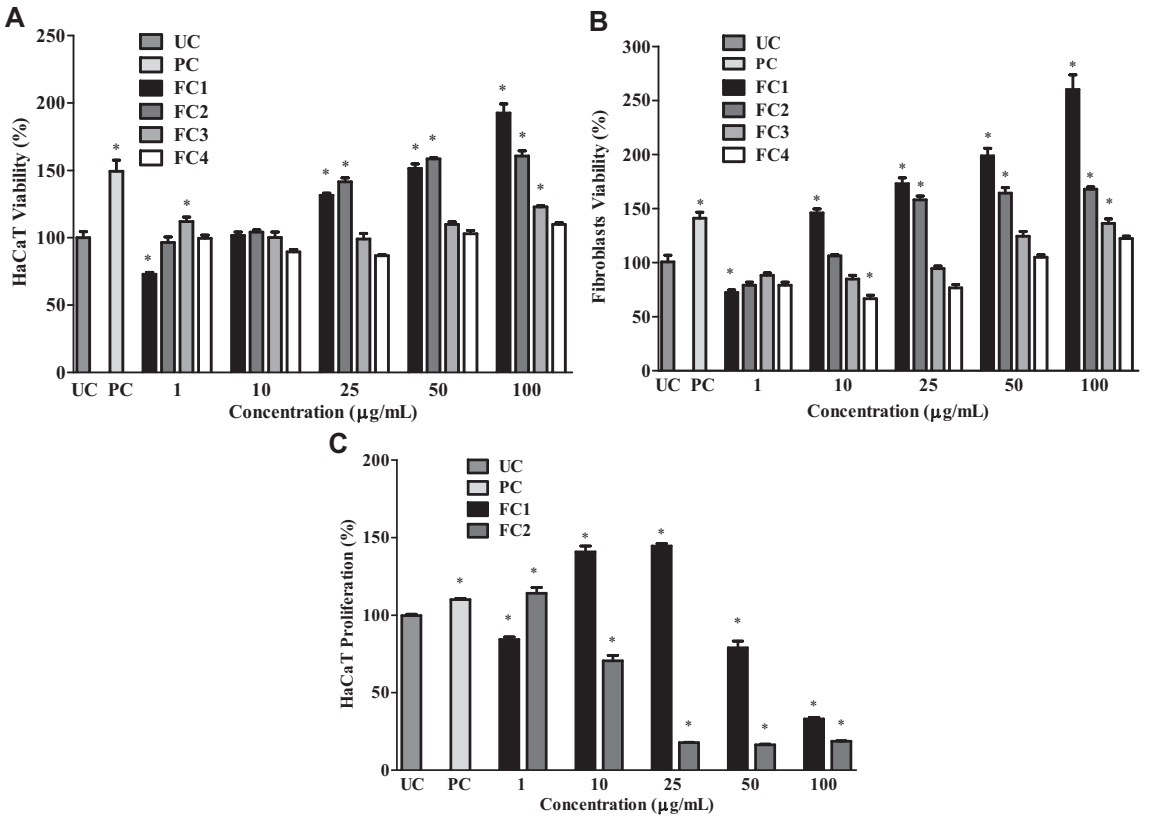

Fig. 2. Influence of FC1, FC2, FC3, and FC4 on cellular dehydrogenase activity (MTT assay) of HaCaT keratinocytes (A) and dermal fibroblasts pNHDF (B). The influence of FC1 and FC2 on mitogenic cell proliferation (BrdU assay) of HaCaT keratinocytes is displayed in (C). The results are expressed as mean ± SD. Values are mean of 3 independent experiments, n = 8 replicates. NC: untreated negative control; PC: positive control 5% FCS; $^{*}p < 0.05$ compared to NC.

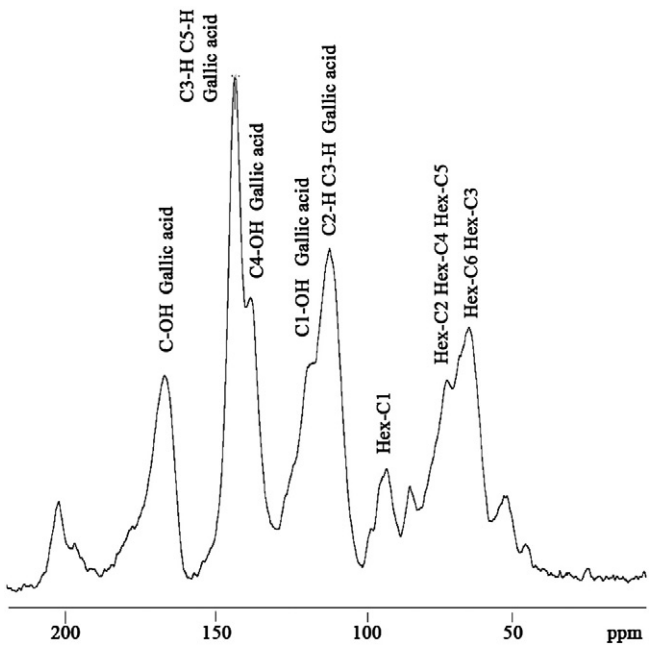

Fig. 3. Solid state ^{13}C NMR spectra of FC1 indicating signals typical for gallic acid and hexose (Hex).

carbons of syringyl units and at 148 ppm, relative to C-3/4 carbons in guaiacyl units) [45].

The ^{13}C NMR spectrum shows the presence of a monomeric sugar between 50 and 100 ppm. C-atoms from hexose (Hex) moiety are characterized by signals at 92 ppm and those from the other five carbons in the region between 62 and 73 ppm: 70 ppm (Hex-C2), 62 ppm (Hex-C3), 70 ppm (Hex-C4), 72.9 ppm (Hex-C5) and 64 ppm (Hex-C6) (Fig. 2) [31,32,44]. The active fraction FC1 was further analyzed by ESI–MS/MS and the structural information was obtained through interpretation of the fragmentation spectra recorded by MS and MS/MS experiments. Table 2 displays the various *m/z* values obtained in the negative ionization mode $[M - H]^-$ and the proposed compounds based on comparison with MS databases and related literature. Fig. 4 shows the structural features of the tentatively identified compounds. Compounds (**1**), (**2**), and (**4**) at *m/z* 125, 169 and 301 were characterized as pyrogallol, gallic acid and ellagic acid, respectively [33–35]. Compound (**3**) was correlated with the structure of gallic acid methyl ester (methyl gallate) with *m/z* 183 and typical fragments observed for subsequent demethylation (*m/z* 169) followed by decarboxylation (*m/z* 125) [33]. Further MS/MS data indicated the presence of ellagitannins and gallotannins. By using the respective MS fragmentation data and published literature the following compounds might be part or present in the fraction, whereby the stereochemical aspects cannot be shown by the MS experiments, but are deduced from already published compounds. *β*-1-*O*-galloyl-3,6-(R)-hexahydroxydiphenoyl-D-glucose (corilagin) at *m/z* 633 (**5**) [36], 1,4,6-tri-*O*-galloyl-glucose at *m/z* 635 (**6**) [37], 1,3-digalloyl-4,6-hexahydroxydiphenoyl-*β*-D-glucopyranoside (tellimagrandin I) at *m/z* 785 (**7**) [38], 1,2,3,6-tetra-*O*-galloyl-glucose at *m/z* 787 (**8**) [37], mallotinic acid at *m/z* 801 (**9**) [39], 1,2,3-trigalloyl-4,6-hexahydroxydiphenoyl-*β*-D-glucopyranoside (tellimagrandin II) at *m/z* 937 (**10**) [40], 1,2,3,4,6-penta-*O*-galloyl-glucose at *m/z* 939 (**11**) [37], geraniin at *m/z* 951 (**12**) [41], and mallotusinic acid at *m/z* 1119 (**13**) [39]. This is the first time that hydrolyzable tannins are described for *P. pluviosa*.

From this point of view FC1 can be assessed as the fraction from *P. pluviosa* responsible for the stimulatory activity of keratinocytes and dermal fibroblasts due to the content of hydrolyzable tannins. Similar ellagitannins (geraniin (**12**), corilagin (**5**), furosin) with strong wound-healing properties have recently been documented as the active compounds in aqueous extracts from *Phyllanthus muellerianus* (Kuntze) Exell [7] with strong stimulating effects against human keratinocytes and dermal fibroblasts. Also for pentagalloylglucose (**11**) reports have been published on potential wound healing properties [4]: this compound was proven to be the main constituent of *Paeonia suffruticosa*, widely used in Traditional Chinese Medicine for its skin activity. Also for trigalloyl-glucose and gallic acid from *Terminalia chebula* in vitro bioactivity against skin cells has been shown [42]. The underlying mechanisms for these stimulatory effects of polyphenols on skin cells are still unclear. On the one side an activation of extramembranous receptors (e.g. KGFR and EGFR) is discussed by the interaction of the polyphenols with the respective proteins of the target cell; similar interactions of galloylated proanthocyanidins with binding pockets of surface receptors have been shown recently [45]. This could finally lead to the increased expression of KGF in keratinocytes, as it has shown recently for the interaction of a polysaccharide with keratinocytes [46]. On the other side differentiation of keratinocytes can be triggered by galloylated flavan-3-ols via the induction of p57/KIP2 pathway [47].

From this point of view the phytochemical composition of FC1 from *P. pluviosa* can easily be correlated towards a rationalized activity for wound healing, also by consideration of the positive outcome of several in vivo studies in rats using polyphenol-enriched extract preparations [43]. From this point of view the traditional use of extracts from *P. pluviosa* stem bark for wound healing seems to be justified [16,17].

4. Conclusions

The results in the present study indicate that hydroalcoholic extract or fraction FC1 from the stem bark of *P. pluviosa* acts as stimulant of human keratinocytes and dermal fibroblasts in vitro. The influence on the cell physiology of skin cells and the pharmacodynamics properties of the extract is due to the high amounts of hydrolyzable tannins. This increase in cellular proliferation of skin cells suggests that epidermal barrier formation can be accelerated by the use of *P. pluviosa* and therefore the extract or the fraction FC1 might be suitable for potential wound healing preparations.

Conflict of interest statement

The authors report no conflict of interest.

Table 2
MS and MS/MS data of compounds detected in subfraction FC1 obtained from the stem bark of *P. pluviosa* and correlation to published data from literature.

Compound	Compound name	Elemental composition	$[M - H]^-$	MS/MS	Literature
1	Pyrogallol	$C_6H_6O_3$	124.9	53, 81, 97, 107	[35]
2	Gallic acid	$C_7H_6O_5$	169.0	53, 81, 97, 107, 125	[34]
3	Gallic acid methyl ester	$C_8H_8O_5$	183.2	78, 95, 124, 168	[33]
4	Ellagic acid	$C_{14}H_6O_8$	301.2	185, 229, 257, 284	[34]
5	Corilagin	$C_{27}H_{22}O_{18}$	633.0	169, 275, 301, 481	[36]
6	1,4,6-Tri-*O*-galloyl- glucose	$C_{27}H_{24}O_{18}$	635.1	125, 169, 221, 271, 313, 423, 465, 483	[37]
7	Tellimagrandin I	$C_{34}H_{26}O_{22}$	785.1	125, 169, 275, 301, 483, 615, 633	[38]
8	1,2,3,6-Tetra-*O*-galloyl-glucose	$C_{34}H_{28}O_{22}$	786.9	125, 169, 295, 403, 421, 447, 465, 617, 635	[37]
9	Mallotinic acid	$C_{34}H_{26}O_{23}$	801.1	301, 463, 633, 757	[39]
10	Tellimagrandin II	$C_{41}H_{30}O_{26}$	936.8	125, 169, 275, 301, 483, 633, 785, 919	[40]
11	1,2,3,4,6-Penta-*O*-galloyl-glucose	$C_{41}H_{32}O_{26}$	938.9	125, 169, 329, 447, 601, 617, 769, 787	[37]
12	Geraniin	$C_{41}H_{28}O_{27}$	951.2	169, 301, 445, 481, 633, 757, 783, 933	[41]
13	Mallotusinic acid	$C_{48}H_{32}O_{32}$	1119.3	125, 169, 301, 445, 633, 757, 801, 933, 951, 1074	[39]

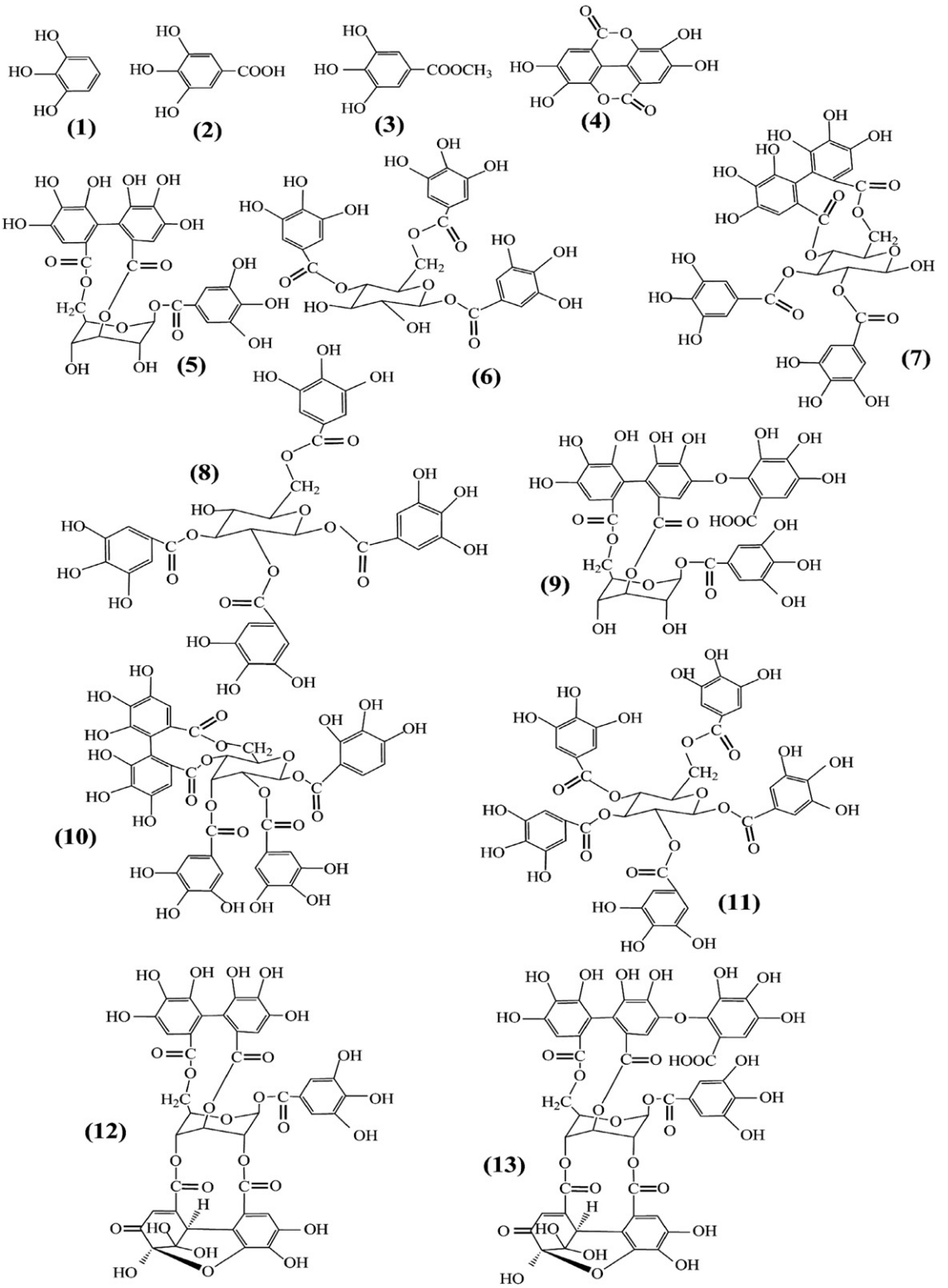

Fig. 4. Structural features of compounds from fraction FC1 from the stem bark of *Poincianella pluviosa*.

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